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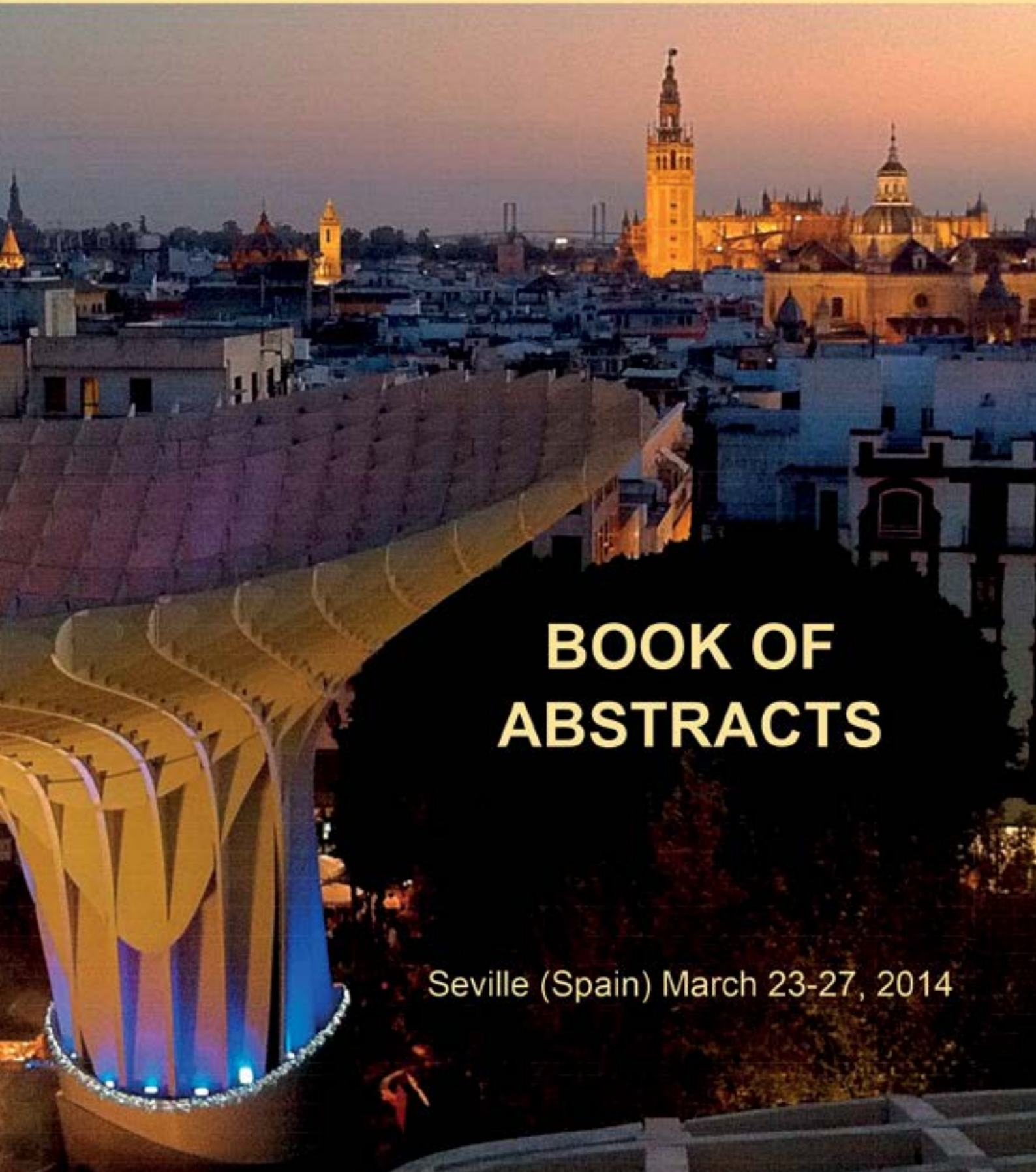

BOOK OF ABSTRACTS

Seville (Spain) March 23-27, 2014

305 SIGNALLING PROCESS AND ACTIVATION OF SLTA, A TRANSCRIPTION FACTOR INVOLVED IN CATION/ALKALINITY STRESS RESPONSE

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Many microorganisms, including fungi, have developed genetic strategies to survive to environment stresses, such as variations in pH, temperature, nutrient availability, reactive oxygen or diverse saline concentrations. In the filamentous fungus and model organism Aspergillus nidulans, tolerance to an alkaline ambient pH requires the activities of three high hierarchy transcription factors: PacC, CrzA and SltA. We have described the role of SltA, a C2H2 zinc-finger transcription factor, in tolerance to alkalinity and to high concentrations of certain mono and divalent cations. Although PacC and CrzA homologues are widely distributed among fungal kingdom, SltA homologues are found only in filamentous fungi. Here we present our latest results in the signalling process and the activation of SltA, in addition to its transcriptional regulatory activity. Signalling of SltA requires its proteolytic processing, an extreme post-translational modification mechanism that shares with PacC. To understand how SltA is signalised and mediates its regulatory action we have isolated mutations affecting this cation/pH response pathway. A source of new slt- mutations was the isolation of extragenic suppressor mutations of the lethal phenotype caused by certain null vps alleles. Several of these mutations mapped in sltA and others allowed the identification of a novel member of this pathway. The new locus has been denoted as sltB. sltB gene encodes for a protein of 1272 amino acids, also specific to filamentous fungi, with two putative functional domains. The N-terminal pseudokinase domain is involved in the proteolysis of native SltA 78 kDa to a 32 kDa form. A second domain is similar to a trypsin-like protease, and our data suggest that SltB is auto-proteolysed through this protease activity. Finally, we have determined that SltB is expressed in a SltA dependent manner. A model of regulation of SltA through SltB activity is presented for this novel cation/alkaline pH regulatory pathway in filamentous fungi.

306 STUDYING THE EXPRESSION OF TERPENE CYCLASE GENES, KEY ENZYMES IN THE SECONDARY METABOLISM OF THE PHYTOPATHOGENIC FUNGUS BOTRYTIS CINEREA

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Botrytis cinerea is a phytopathogenic fungus that shows a wide variety of mechanism for infecting plant material. B. cinerea has a complex secondary metabolism, allowing infecting more than two hundreds plant species. Until date, several families of cell wall degrading enzymes and toxins have been described for this fungus1, including two important families of toxins: the sesquiterpene botrydial (and related compounds), and botcinic acid and its derivated2,3. In the genome of this fungus, six putative sesquiterpene cyclases and three diterpene cyclases genes have been annotated1, but not all of them have been totally characterized yet. It is known that STC1 encodes for a sesquiterpene synthase (BcBOT2) which plays a crucial role in the biosynthesis pathway of toxin botrydial3. The biological role of the other STC and DTC genes is currently being elucidated.

This study presents the analysis of expression profiles shown by quantitative reverse transcription-PCR of the terpene gene family (STC and DTC) encoding sesquiterpene cyclase and diterpene enzymes in B. cinerea. We used an OSMAC approach (one strain many compounds) trying to stimulate the secondary metabolisms of this fungus, and we described the evolution of the gene expression profiles during several days of fermentation.

1.- Amselem J, Cuomo CA, van Kan JAL, Viaud M, Benito EP, et al. (2011) Genomic Analysis of the Necrotrophic Fungal Pathogens Sclerotinia sclerotiorum and Botrytis cinerea. PLoS Genet 7(8): e1002230. doi:10.1371/journal.pgen.1002230

2.- Collado IG, Macias Sánchez AJ, Hanson JR. (2007) Fungal terpene metabolites: biosynthetic relationships and the control of the phytopathogenic fungus Botrytis cinerea. Natural Product Report, 24(4), 674-686.

3.- Pinedo C, Wang CM, Pradier JM, Dalmais B, Choquer M, Le Pêcheur P, Morgant G, Collado IG, Cane DE, Viaud M. (2008) Sesquiterpene synthase from the botrydial biosynthetic gene cluster of the phytopathogen Botrytis cinerea. ACS Chemical Biology, 3(12), 791-801.

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